

## AMENDMENTS TO THE CLAIMS:

### Listing of Claims:

1. (Currently amended): A method of separating sperm cells, comprising:
  - [[a.]] obtaining semen from a male of a species of mammal which contains a plurality of sperm cells;
  - [[b.]] incubating said semen at a temperature above which sperm cell membrane lipids transition from a liquid phase to gel phase;  
staining said sperm cells with Hoechst 33342 stain for a period of 30 minutes;
  - [[c.]] determining a ~~sperm-cell~~ sex characteristic of a plurality of said sperm cells;
  - [[d.]] separating said sperm cells based upon said ~~sperm-cell~~ sex characteristic into X chromosome bearing and Y chromosome bearing populations; and
  - [[e.]] collecting separated sperm cells.
- 2-3. (Cancelled)
4. (Original): A method of separating sperm cells as described in claim 1, wherein said step of incubating said semen at a temperature above which sperm cell membrane lipids transition from a liquid phase to gel phase temperature comprises incubating said semen at a temperature between about 5° C and about 25° C.
5. (Original): A method of separating sperm cells as described in claim 1, wherein said temperature is selected from the group consisting of about 5 ° C, about 6° C, about 7° C, about 8° C, about 9° C, about 10° C, about 11° C, about 12° C, about 13° C, about 14° C, about 15° C, about 16° C, about 17° C, about 18° C, about 19° C, about 20° C, about 21° C, about 22° C, about 23° C, about 24° C, and about 25° C.

6. (Original): A method of separating sperm cells as described in claim 1, wherein said species of mammal is selected from the group consisting of a bovine species of mammal, an equine species of mammal, an ovine species of mammal, a swine species of mammal, a canine species of mammal, a feline species of mammal, a deer species of mammal, an elk species of mammal, and a marine species of mammal.
7. (Original): A method of separating sperm cells as described in claim 1, wherein said species of mammal comprises a bovine species and wherein said step of incubating said semen at a temperature above which sperm cell membrane lipids transition from a liquid phase to gel phase temperature comprises incubating said semen at a temperature between about 17° C and about 19° C.
8. (Original): A method of separating sperm cells as described in claim 1, wherein said species of mammal comprises a bovine species and wherein said step of incubating said semen at a temperature above which sperm cell membrane lipids transition from a liquid phase to gel phase temperature comprises incubating said semen at a temperature of about 17° C.
9. (Original): A method of separating sperm cells as described in claim 1, wherein said species of mammal comprises an equine species and wherein said step of incubating said semen at a temperature above which sperm cell membrane lipids transition from a liquid phase to gel phase temperature comprises incubating said semen at a temperature of about 15° C.
10. (Original): A method of separating sperm cells as described in claim 1, wherein said step of incubating said semen at a temperature above which sperm cell membrane lipids transition from liquid phase to gel phase comprises incubating said semen at said temperature above which sperm cell membrane

lipids transition from liquid phase to gel phase between about one hour to about 18 hours.

11. (Original): A method of separating sperm cells as described in claim 1, further comprising the step of transporting said semen from a first location to a second location during said step of incubating said semen at a temperature above which sperm cell membrane lipids transition from a liquid phase to gel phase.

12. (Original): A method of separating sperm cells as described in claim 1, further comprising the step of adding an antibacterial to said semen prior to said step of incubating said semen at a temperature above which sperm cell membrane lipids transition from a liquid phase to gel phase.

13-14. (Cancelled)

15. (Currently amended): A method of separating sperm cells as described in claim 1, further comprising the step of extending semen with an extender ~~selected from the group consisting of KMT, and INRA96.~~

16. (Original): A method of separating sperm cells as described in claim 1, further comprising the step of concentrating said sperm cells by removing a portion of seminal plasma.

17. (Cancelled)

18. (Currently amended): A method of separating sperm cells as described in ~~claim 17~~ claim 1, wherein said step of staining said sperm cells comprises staining DNA contained within said sperm cells.

19-20. (Cancelled)

21. (Original): A method of separating sperm cells as described in claim 1, wherein said step of separating said sperm cells based upon said sperm cell characteristic comprises separating said sperm cells using an instrument selected from the group consisting of a flow cytometer, and a cell sorter.
- 22-147. (Cancelled)
148. (New): A method of separating sperm cells as described in claim 1, wherein said step of incubating said semen at a temperature comprises incubating said semen at 15°C and further comprising the step of using said separated X chromosome bearing sperm cell population or said separated Y chromosome sperm population to provide about 72% pregnancy rate.
149. (New): A method of separating sperm cells as described in claim 1, and further comprising the step of using said separated X chromosome bearing sperm cell population or said separated Y chromosome sperm population with hysteroscopic insemination.
150. (New): A method of separating sperm cells as described in claim 15 wherein said extender is selected from a group consisting of Tyrode's-based skim milk-glucose extender, and Kenney modified Tyrodes.
151. (New): A method of separating sperm cells as described in claim 1 and further comprising the step of adding caffeine to said semen.
152. (New): A method of separating sperm cells as described in claim 1 wherein said step of staining said semen with Hoechst 33342 comprises the step of staining  $50 \times 10^6$  sperm/mL of semen with a concentration of Hoechst 33342 between about 2.6  $\mu$ l and about 3.9  $\mu$ l.

153. (New): A method of separating sperm cells as described in claim 1 wherein said step of staining said semen with Hoechst 33342 comprises the step of staining between  $150 \times 10^6$  sperm/mL and  $450 \times 10^6$  sperm/mL of semen with a concentration of Hoechst 33342 between about 7.8  $\mu$ l and about 23.4  $\mu$ l.
154. (New): A method of separating sperm cells as described in claim 1 and further comprising the step of providing a pH of a stain solution between about 7.2 pH to about 8.0 pH.